Comparative Fine Structure of Acarine Integument[1]

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Abstract: The integument shows a fundamental similarity in representatives of three suborders of mites considered in this work, despite variation in thickness and variety of organ types present.

After aldehyde fixation and mixed-resin embedding, use of the electron microscope revealed distinct divisions of the mite cuticle. A widely laminated endocuticle overlies the hypodermis, a heavily sclerotized exocuticle is present, and non-laminated epicuticle appears to be deposited outermost. Pore canals and ducts with fixed hypodermal secretions are obvious in several sections. Setae with sockets and sense organs are found at intervals in the cuticle.

Many unanswered questions exist on the subject of acarine integument, especially the structural differences in the cuticle of mites in the various suborders. Using specific techniques, the electron microscope disclosed details of integumental structures which amplify current information about the integument in relation to classification and function.

Mites from three suborders of the Acarina were considered: a) *Parasitus sp.* from the Mesostigmata, b) *Tydeus sp.* from the Prostigmata, and c) *Oppia sp.* from the Cryptostigmata. All mites sectioned were adults. Although adults are more difficult to section because of a harder cuticle, one can be sure that all formation of the integument has been completed. The posterior half of the mites was used in the sectioning because in each of the specimens used the anterior portions are more contoured and more unevenly sclerotized.

MATERIALS AND METHODS

A solution of 6.25% buffered glutaraldehyde provided rapid and adequate fixation for entire, living mites immersed directly into the fixative. Another fixative used quite successfully was a mixture of picric acid and paraformaldehyde. After fixing the specimens for approximately four hours, a post fixative of 1% phosphate-buffered osmium tetroxide (Millonig, 1961) was employed for about thirty minutes.

After fixation and rinsing in phosphate buffer, the specimens were dehydrated in a graded series of ethyl alcohols. Insufficient dehydration experienced in earlier procedures may have been caused by the sclerotized exoskeleton of the mites which allowed the water-alcohol exchange to progress slowly. Conse-

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quently, fifteen minute changes in the 35%–95% range, and two changes of 20–30 minutes for the absolute alcohol were necessary.

Propylene oxide proved to be the best transitional solvent. This solvent, combined with an embedding mixture of Epon 812 (Shell Chem. Corp., San Francisco, Calif.) and Araldite 502 (Ladd Research Industries, Burlington, Vt.), allowed sufficient infiltration of plastic into the specimen. The resin mixture consisted of one part Epon, one part Araldite, 1.5 parts DDSA (Shell Chem. Corp.), with 1.5% catalyst (Geisy, 1967). The fixed and dehydrated mites were passed through two changes of pure propylene oxide, and placed in a mixture of 3 : 1 concentration of solvent to resin for one hour; then a 2 : 2 concentration for one hour; and one part solvent to three parts resin mixture for at least fifteen hours. Specimens were then changed into solutions of pure resin for twenty-four hours at room temperature. A final change into fresh resin was made in an oven at 60°C for 2–3 days.

Sections were cut with glass knives on an LKB 'Ultratome' and Porter-Blum microtome. Usable sections varied from gold to gray in color and were probably 400–900 Å thick. Tissues were stained with uranyl acetate and lead citrate.

OBSERVATIONS AND RESULTS

Parasitus sp. (Parasitidae), Figs. 1 and 2

This amber colored mite is relatively large (800–1000μ); it is predaceous on small insects and other mites, and was collected in Colorado from predominantly mesophyllic habitats of pine and leaf litter. The thickness of the integument varies from 7–9 microns depending upon the area sectioned.

The most outstanding characteristic of the integument is the distinct laminations (Fig. 1). These laminations vary in thickness depending upon the area of integument. They are usually seen in electron micrographs as alternating light and dark bands, and appear narrower and more compressed toward the epicuticle (Fig. 1). This difference in laminar structure is explained by Richards (1951). The laminae appear to shrink considerably in thickness after formation. This is caused by sclerotization and darkening of the older cuticle due to some dehydration with subsequent increase in density, resulting in the appearance of the tightly packed laminations in the exocuticle.

The epicuticle appears to be a single, non-laminated layer, overlying the exocuticle (Fig. 1). In the mites the epicuticle is a waxy layer, probably similar to that reported by Beament (1968). In this case, it has been partially removed by dissolution in one of the preparatory solutions.

Pore canals are minute ducts extending upward from the hypodermis through the endocuticle and exocuticle (Fig. 1). They are quite small (½–1μ in diameter) and can hardly be resolved as ducts by the light microscope. These pores can be seen with the electron microscope. The entire cuticle of this Parasitidae mite appears replete with pore canals. The helical nature of the

FIG. 1. Cross section through the integument of a *Parasitus sp.* mite showing laminations (LAM), divisions of the cuticle as endocuticle (ENC), exocuticle (EXC), and epicuticle (EPC), and pore canals (PC) containing secretory material. × 15,000.

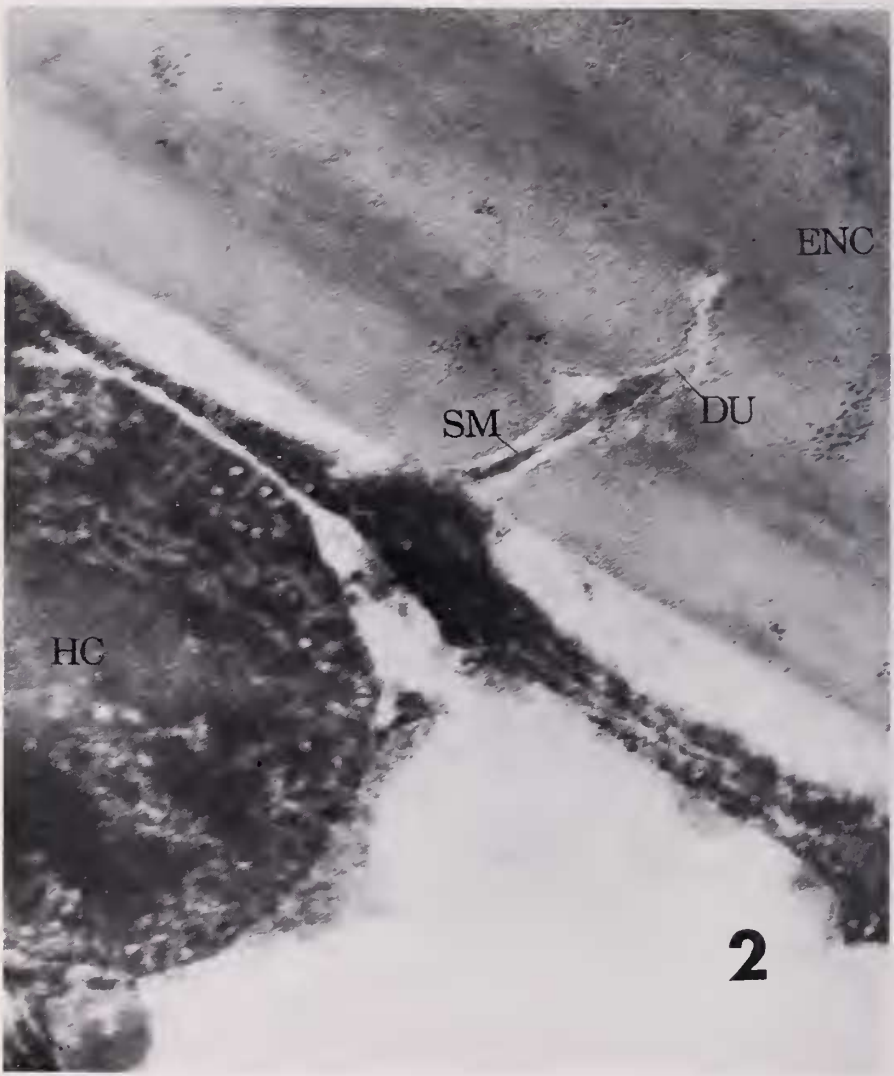

FIG. 2. Enlargement of a portion of the endocuticle (ENC) of a parasitidae mite. A duct (DU) filled with secretory material (SM) runs into the endocuticle. Portion of a hypodermal cell (HC). × 39,000.

canals is ascertained by noting their tortuous routes and intermittent areas of lighter density where the microtome knife has shaved only portions of a single, twisting pore canal (Fig. 1). The large number of pore canals suggests the constant production of waxy secretions as an outermost protective epicuticle.

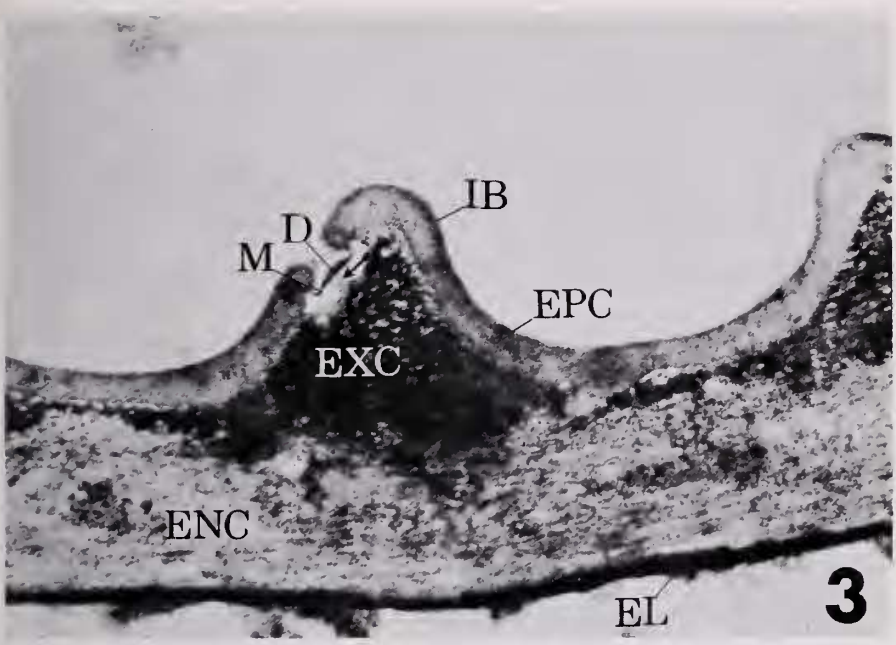

Fig. 3. Cross section through the integument of a *Tydeus sp.* mite showing distinct divisions of the cuticle as endocuticle (ENC), exocuticle (EXC), and epicuticle (EPC). Components of the "campaniform sensillus" are labeled membranous portion (M), dome portion (D), and innervating fiber (arrow). Integumental bump (IB), electron dense layer (EL). × 14,500.

The general hypodermal cells are known to secrete the cuticle (Condoulis and Locke, 1966) probably manufacturing a part of its constitutents. In this mite, the hypodermis is typically a single layer of cells, each cell with a rather extensive endoplasmic reticulum. Under high magnification, hypodermal secretions can be seen in ducts which traverse the integument (Fig. 1 and 2).

Tydeus sp. (Tydeidae), Figs. 3 and 4

This mite is rather small (200–300μ); it is generally predaceous on small insects and other mites. It has been collected from practically every habitat in Colorado where mites are found and is easily noticed because of its attractive yellow, pink, or red integument. The cuticle of this mite shows a great variety in thickness, but is generally 2–4 microns thick in cross section.

In comparison to the *Parasitus sp.* mite, the integument of this mite can hardly be termed laminated, but shows a definite endocuticle, exocuticle, and epicuticle (Fig. 3).

The endocuticle appears very lightly sclerotized, unordered, and even fibrous in some cases. The exocuticle, although more heavily sclerotized, is also un-

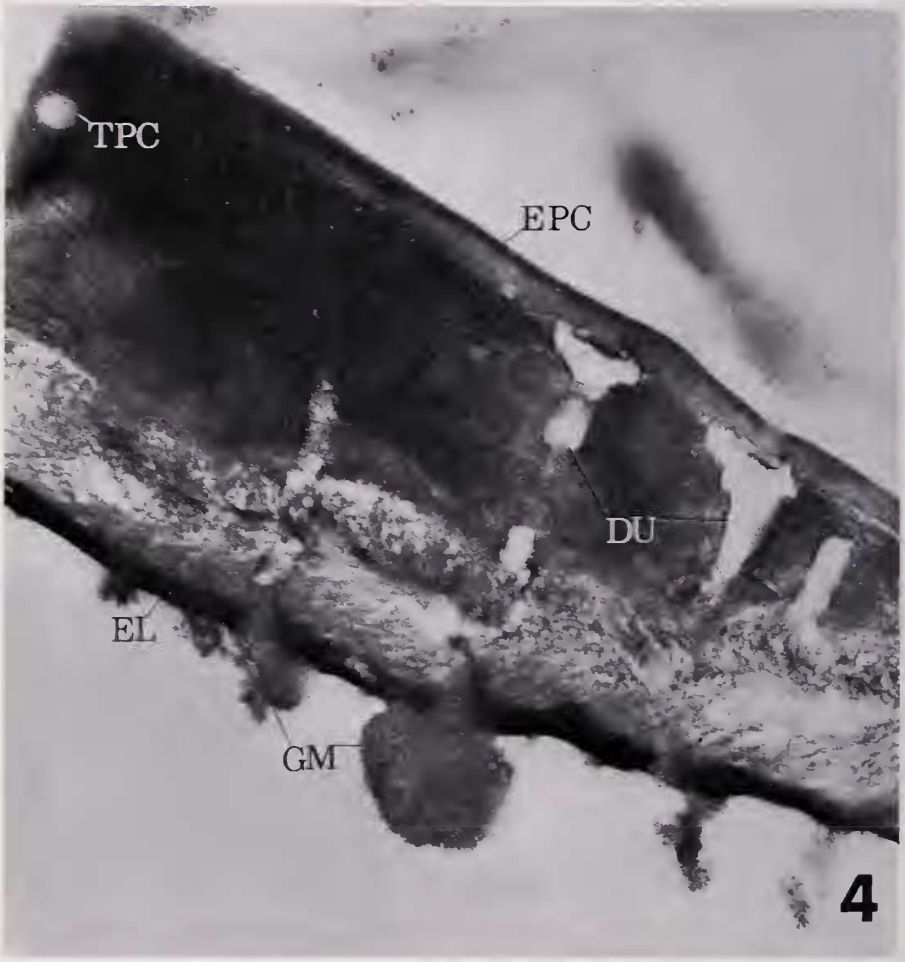

FIG. 4. Enlargment of a cross section through the integument of a tydeidae mite. Glandular material (GM) is fixed in several ducts (DU) which may branch below the epicuticle (EPC). Electron dense layer (EL), transverse pore canal (TPC). × 38,000.

ordered and fibrous. The epicuticle is unusually thick in this species. The waxy epicuticle appears as though it would provide adequate protection for the softer, fibrous portions of the cuticle below. A heavy, electron-dense layer separates the endocuticle from the underlying hypodermis (Figs. 3 and 4).

Glandular material in association with ducts is definitely visible (Figs. 2, 4, and 5). The secretion can be seen in the canals and was apparently fixed while moving toward the epicuticle. In several cases, the ducts branch just below the epicuticle (Fig. 4), and I infer that they pour glandular secretions directly

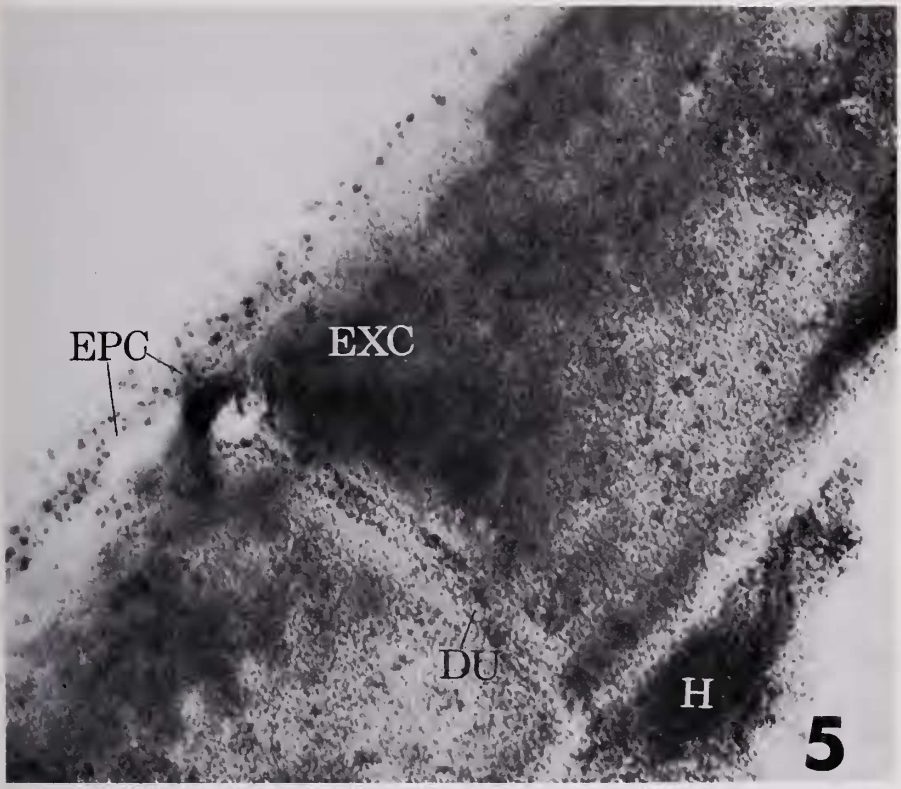

Fig. 5. Cross section through the integument of an *Oppia sp.* mite demonstrating formation of the epicuticle (EPC) after the lipid substance has emerged from a duct (DU) of the hypodermis (H) and spread over the surface of the exocuticle (EXC). × 45,500.

onto the surface of the exocuticle (Fig. 5). This situation is similar to that reported by Locke (1959, 1961, 1965) in several insects. Similar ducts have previously been reported by Wharton *et al.* (1968) in the mite *Laelaps echidnina*.

One of the more distinguishing characteristics for identifying the Tydeidae by light microscopy is a dotted integumental pattern on the dorsum of the mite. Electron microscopy has revealed the apparent reason for this pattern as a regular series of bumps and depressions which correspond respectively to the light and dark areas of the dotted design seen under the light microscope (Fig. 3). On one of these integumental bumps is a "campaniform sensillus" as described by Wigglesworth (1965). This is a sensory receptor which is thought to respond to air pressure or vibrations, and possibly to bending of the cuticle. The receptor consists of a dome portion, a thin membrane, and an innervating fiber (Fig. 3). The latter is evidence that this is a sensory structure. Because the epicuticle is interrupted in an orderly arrangement, I infer that this is not an artifact.

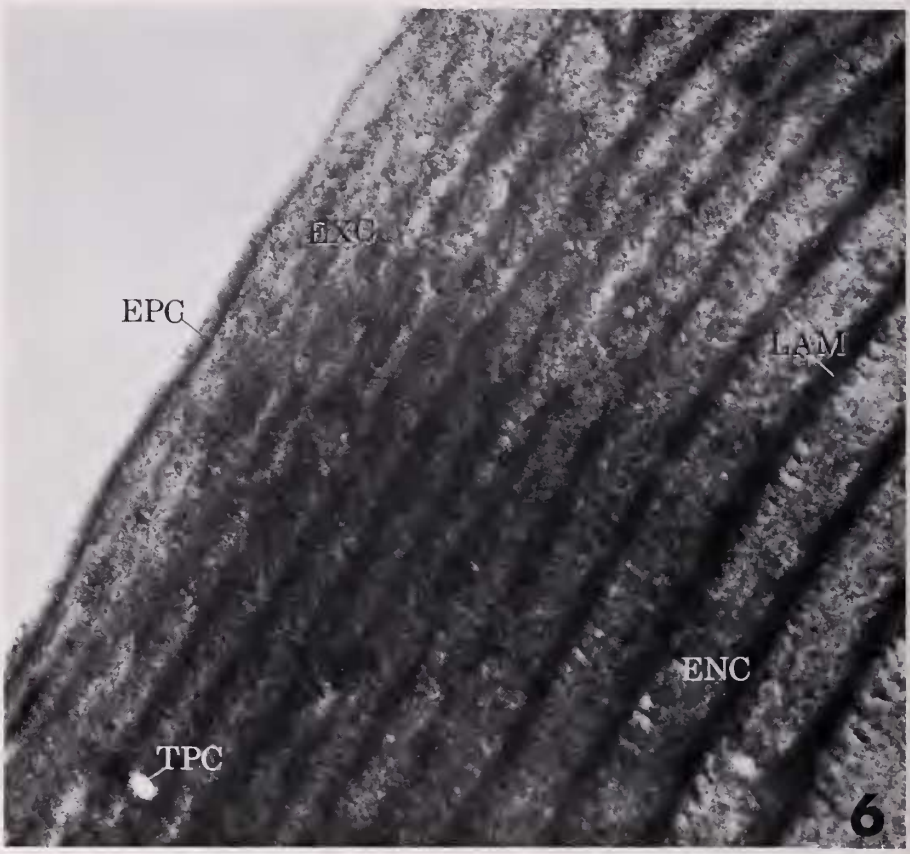

FIG. 6. Cross section through the integument of an oppiidae mite showing the laminations (LAM), the endocuticle (ENC), the compact exocuticle (EXC), and the waxy epicuticle (EPC). Transverse pore canal (TPC). × 34,000.

Oppia sp. (Oppiidae), Figs. 5, 6, 7, 8, and 9

This mite is of moderate size (400–600μ). It was collected from both hygrophyllic and mesophyllic habitats, and was raised successfully in the laboratory on a diet of fermented yeast and dextrose (Dolan, 1968).

Most oribatid mites have a heavily sclerotized and highly laminated cuticle. This oppiid is no exception (Fig. 6). Cuticle thickness varied between five and seven microns. The endocuticle and exocuticle are both laminated, but the laminae in the exocuticle are more tightly packed and not so well defined. The non-laminated epicuticle lies outermost. Transverse pore canals are quite common (Fig. 6), and are not confined to any particular area of the integument.

Setae are typically found on these and most other mites. Even though Wigglesworth (1965) has described the basic structure and formation of a typical seta,

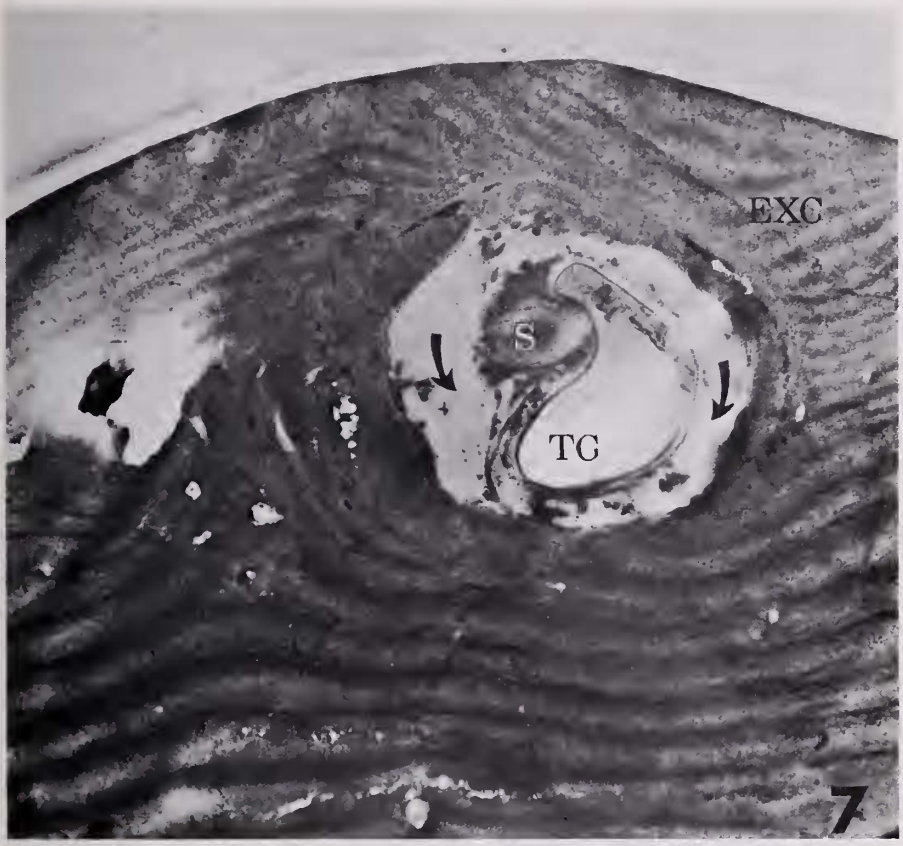

Fig. 7. Cross section through the integument of an oppiidae mite revealing a setal complex. The solid seta (S) and its tormogen cell (TC) are located in a socket filled with embedding plastic (arrows). Exocuticle (EXC). × 12,000.

nothing has been reported in the literature on its fine structure. Considering this, several pertinent points are deducible from Fig. 7. The seta is solid throughout. The tormogen or socket-forming cell lies in close proximity. Judging from the amount of embedding plastic which surrounds the seta and its tormogen cell (Fig. 7), it can be assumed that the complex is nestled in an open follicle. Note how the integument has compensated for the presence of a seta by the divergence of the laminar material around the follicle (Fig. 7).

There appears to be another form of "campaniform sensillus" in the integument of this mite (Fig. 8). Again note the dome portion, membranous folds, and obvious innervating fiber, all described as parts of a sensory receptor of this kind.

Fig. 8. A form of "campaniform sensillus" in the cuticle of an oppiide mite. Membranous portion (M), dome portion (D), innervating fiber (arrow). × 43,000.

The muscles of arthropods are united to the cuticle in several different ways (Richards, 1951). A predominant feature of the Cryptostigmata are apodemata, heavily sclerotized infoldings of the cuticle which provide areas for muscle attachment. Distinct rugosites are found on the interior surface of this apodeme of the ventral plate, and muscles are attached to them (Fig. 9).

SUMMARY

The laminated integument of mites is secreted by the hypodermal cells. Minute pore canals and larger ducts extend from the hypodermis, traverse the integument, and ultimately carry secretory material through the endocuticle to the surface of the exocuticle. These waxy secretions solidify as the protective epicuticle. Beament (1959) described the removal of this covering from various arthropods by abrasion, melting, and dissolution. These renovations exposed openings of numerous pore canals to the exterior, and demonstrated the protective function of the epicuticle since dehydration of the test animals ensued.

The electron microscope has revealed some of the fine structure of several cuticular elements. The large seta on the dorsum of an oribatid mite is nestled in a socket complete with a tormogen cell. In addition, the pressure receptors of the cuticle appear dependent on the stretchable membranous portions that

Fig. 9. This cross section through an apodeme of the ventral plate (AVP) of an oppiid mite shows rugosities (R) for muscle (M) attachment. Ventral plate (VP). × 12,000.

probably relay messages through the dome and inervating fiber when the cuticle is bent.

Literature Cited

Beament, J. 1959. The waterproofing mechanism of arthropods. J. Exp. Biol., **36:** 391–422.

———. 1968. The insect cuticle and membrane structure. British Med. Bull., **24:** 130–134.

Condoulis, W. and M. Locke. 1966. The deposition of endocutcile in an insect, *Calpodes ethlius* (Lepidoptera, Hesperiidae), J. Insect Physiol., **12:** 311–323.

Dolan, J. 1968. Personal communication. Department of Zoology, Colorado State University, Fort Collins, Colorado.

Geisy, R. 1967. Personal communication. Department of Botany and Plant Pathology, The Ohio State University, Columbus, Ohio.

Locke, M. 1959. Secretions of wax through the cuticle of insects. Nature, **184:** 1967.

———. 1961. Pore canals and related structures in insect cuticle. J. Biophys. Biochem. Cytol., **10:** 589–618.

———. 1965. Permeability of insect cuticle to water and lipids. Science, **147:** 295–298.

Millonig, G. 1961. Advantages of a phosphate buffer for osmium tetroxide solutions in fixation. J. Appl. Phys., **32:** 1637.

Richards, A. 1951. The integument of arthropods. Univ. of Minn. Press, pp. 18–95.

Wharton, G., W. Parrish and D. Johnston. 1968. Observations on the fine structure of the cuticle of the spiny rat mite *Laelaps echidnina* (Acari-Mesostigmata). Acarologia, **10:** 206–214.

Wigglesworth, V. 1965. The principles of insect physiology. Methuen & Co., London, England, pp. 25–272.